

Proceedings of the Annual International Conference on Soils, Sediments, Water and Energy

Volume 12

Article 13

January 2010

Mathematical Modeling And Experimental Studies On Biochemical Conversion Of Cr(Vi) Of Tannery Effluent To Cr(Iii) In A Chemostat

K. Samanta

Jadavpur University

P. Bhattacharya

Jadavpur University

R. Chowdhury

Jadavpur University

Follow this and additional works at: <https://scholarworks.umass.edu/soilsproceedings>

Recommended Citation

Samanta, K.; Bhattacharya, P.; and Chowdhury, R. (2010) "Mathematical Modeling And Experimental Studies On Biochemical Conversion Of Cr(Vi) Of Tannery Effluent To Cr(Iii) In A Chemostat," *Proceedings of the Annual International Conference on Soils, Sediments, Water and Energy*: Vol. 12 , Article 13.

Available at: <https://scholarworks.umass.edu/soilsproceedings/vol12/iss1/13>

This Conference Proceeding is brought to you for free and open access by ScholarWorks@UMass Amherst. It has been accepted for inclusion in Proceedings of the Annual International Conference on Soils, Sediments, Water and Energy by an authorized editor of ScholarWorks@UMass Amherst. For more information, please contact scholarworks@library.umass.edu.

Chapter 12

MATHEMATICAL MODELING AND EXPERIMENTAL STUDIES ON BIOCHEMICAL CONVERSION OF CR(VI) OF TANNERY EFFLUENT TO CR(III) IN A CHEMOSTAT

K. Samanta, P. Bhattacharya, and R. Chowdhury

Chemical Engineering Department, Jadavpur University, Kolkata-700032, India

Abstract: Biodegradation of hexavalent chromium present in tannery waste has been studied using *Pseudomonas* sp. (JUBTCr1) and *Bacillus* sp. (JUBTCr3) isolated from their native source. A 5L double-jacketed chemostat with 4L working volume has been used as contacting device for the kinetic investigation of the biodegradation process. Varying the feed volumetric rate from 118-133 ml/h for different inlet hexavalent chromium concentrations (30 to 90 mg/dm³), an attempt has been made to study the reaction engineering behavior of the system. It is observed that Haldan-type substrate inhibited model can satisfactorily be used to predict the extent of bioconversion for different dilution rate. Using the kinetic parameters of proposed Haldane equation, a CSTR model was developed. Model prediction agreed well with experimental data.

Key words: Tannery waste; Hexavalent chromium removal; Microbial route; Growth kinetics; Haldane type inhibition, mathematical-modeling.

1. INTRODUCTION

Hexavalent chromium (Cr⁺⁶) is often found in soil and ground water due to its wide spread industrial use in several process industries (Ganguly and Tripathi, 2002, Stern, 1982), such as tannery, electroplating, steel industries etc. Cr⁺⁶ are highly toxic (Zhang et al., 2001, Flores and Perez, 1999), carcinogenic (Lurie and Wolfe, 2002, Losi et al., 1994) and mutagenic (Brien et al., 2001, Stearns et al., 1995) pollutant even at very low concentration (Venitt and Levy, 1974, EPA, 1998, McLean and Beveridge, 2000). As many aerobic and anaerobic microorganisms are capable of reducing Cr⁺⁶ to Cr⁺³, bioremediation may play an important role for the detoxification from Cr⁺⁶ even at very low (ppm or ppb) level.

It has already been reported that due to the presence of some enzymes called chromium reductases (Gu and Cheung, 2001), different microorganisms belonging particularly to the genus, *Pseudomonas* can reduce Cr⁺⁶ to Cr⁺³. The reduction of transformation capacity of Cr⁺⁶ by microorganisms at higher initial concentration of Cr⁺⁶ has been observed by other researchers (Arellano et al., 2004, Middleton et al., 2003) and the phenomenon has been explained by the presence of inhibitory effect of Cr⁺⁶ at high concentration level (Turick et al, 1997). A detailed literature search reveals that although a considerable quantum of investigation has been reported on bioremediation techniques, a meaningful bioprocess study is still awaited.

In the present investigation bacterial strains, namely, *Pseudomonas* sp. and *Bacillus* sp isolated from tannery effluent, have been used for the microbial transformation of hexavalent chromium to trivalent one. Experimental studies have been conducted in batch mode to obtain different intrinsic kinetic parameters of growth of Cr⁺⁶-reducing bacteria. Simultaneously treatment of tannery waste has been done using the same microbial strains in a chemostat with an intension to achieve the high bioconversion. The initial Cr⁺⁶ concentrations in the effluent and the dilution rate have been

considered as process parameters. Starting from differential mass balance, judiciously combined with the Haldane equation, a semi-deterministic mathematical model equation has been developed which is capable of predicting the extent of bioconversion as a function of dilution rate in the chemostat. The simulated equation contains no adjustable parameter and the constants have been computed. The experimental data obtained through a comprehensive and programmed study have been used to verify the proposed equation.

2. MATERIALS AND METHODS

2.1 Materials

2.1.1 Bacterial strains

Microorganisms, namely, *Pseudomonas* sp. and *Bacillus* sp., isolated and purified from polyculture, collected from the liquid effluent of local tanneries, situated in Kolkata, India, has been used for the present investigation.

2.1.2 Chemicals

NH₄Cl (Ranbaxy), MgSO₄, 7H₂O (E. Merck), FeSO₄, 7H₂O (E. Merck), CaCl₂, 2H₂O, (E. Merck), CH₃COONa, 3H₂O (E. Merck), K₂HPO₄ (E. Merck), Yeast extract powder (LOBA chemie), S-diphenylcarbazide (E. Merck), Acetone, AR (E. Merck), H₂SO₄ (E. Merck), MOPS-NaOH buffer (Sigma), K₂Cr₂O₇ (E. Merck) have been used.

2.2 Analytical Instruments

Electronic balance (Afcoset FX-300), B.O.D Incubator-shaker (S.C.Dey & Co., Kolkata), Laminer flow (Bhattacharya & Co., Kolkata), Cold centrifuge (Remi Instrument Pvt. Ltd., Mumbai), Hot air oven (Bhattacharya & Co., Kolkata), Autoclave (G.B. Enterprise, Kolkata), UV shower (Bhattacharya & Co., Kolkata), Furnace (Bhattacharya & Co., Kolkata), Double beam UV-VIS spectrophotometer (Chemito) have been used.

2.3 Methods

2.3.1 Bacterial enrichment

Isolation of monoculture of the individual genus of microorganisms and their subsequent enrichment has been carried out in Lurea broth (acetate-minimal medium) (Pattanapipitpaisal, 2001a, 2001b) composed of (per litre) NH₄Cl (1 g), MgSO₄, 7H₂O (0.2 g), FeSO₄, 7H₂O (0.001 g), CaCl₂, 2H₂O (0.001 g), CH₃COONa, 3H₂O (5 g), K₂HPO₄ (0.5 g) and yeast extract powder (0.5 g)). The phosphate source was autoclaved separately in 10 ml distilled water. It was cooled and then added to the rest of the medium. Initially the inoculum of each bacterium (1ml) was inoculated in 20 ml of sterile (0.2 M.Pa., 121⁰C, 15 min) selective media. The growth medium was supplemented with K₂Cr₂O₇ solution at a concentration of 1 mg /dm³ of Cr⁺⁶.

Adaptation of the monoculture consortium to the new microenvironment was carried out for seven days by incubating at 28⁰C in a rotary shaker at 150 rpm. Enriched culture was obtained by repeated inoculation of preceding bacterial culture in fresh selective medium containing K₂Cr₂O₇.

2.3.2 Batch Experiments

To determine the growth kinetics of the microorganisms with respect to different initial Cr⁺⁶ concentration in culture medium, batch experiments (Turick et al, 1997, Bajt et al., 1993) were

conducted in Erlenmeyer flasks with constant shaking. For each run the working hold up volume was maintained at 20 dm³ containing 10 % of inoculum. A constant temperature of 32°C was maintained. Samples were withdrawn at an interval of 4 hours with a total time span of 48 hours. Two separate batch experiments were conducted with different initial Cr⁺⁶ concentration ranging from 30 mg/dm³ to 90 mg/dm³.

2.3.3 Chemostat

A 5 dm³ double-jacketed Continuous stirred tank reactor (CSTR), fitted with a mechanical agitator was filled with 4 dm³ of sterile tannery effluent. The reactor content was initially inoculated with 0.4 dm³ of inoculum and kept for 10 hours at an ambient temperature of 30°C to allow the cell growth to reach the exponential phase. A stirrer speed of 200 rpm was maintained. The individual feed solution was then sent continuously to the chemostat through a rotameter. The content was well stirred at 200 rpm and the temperature was maintained at 30°C. The products were withdrawn continuously. For each initial Cr⁺⁶ concentration, the volumetric flow rate of the feed was varied from 117-137 dm³/hr and steady state was allowed to attain. Liquid effluent of the reactor was analyzed after the attainment of the steady state.

The Cr⁺⁶ concentration in different feed solutions was varied from 70 mg/dm³ to 90 mg/dm³ in the chemostat studied.

2.3.4 Determination of Biomass Concentration

The concentration of bacterial mass in the reaction broth of batch type experiments and the CSTR under steady state was determined both spectrophotometrically and by dry weight method (Shuler, and Kargi, 1992). In this method 20 ml nutrient broth, enriched with bacterial strains, was centrifuged at the rate of 10,000 rpm for 15 minutes and the bacterial mass was washed with buffer solution. The washed wet cell mass was then transferred to a pre-weighed aluminum cup and was dried at 80°C for 24 hours. The exact weight of the bacterial mass was determined by subtracting the weight of dry cup from that of the cup containing dry bacterial mass. Cell concentration of bacterial suspensions withdrawn at different time intervals was also determined at 600 nm, spectrophotometrically.

2.3.5 Hexavalent Chromium Analysis

The sample under investigation was centrifuged at 10,000 rpm for 15 minutes and the supernatant was collected to measure the residual Cr⁺⁶ concentration. The analysis of Cr⁺⁶ concentration was carried out spectrophotometrically using Cr⁺⁶ specific colorimetric reagent S-diphenylcarbazide (Pattanapitpaisal, 2001a, 2001b). Initially 0.025 g S-diphenylcarbazide was dissolved in 9.67 ml acetone (AR) to prepare S-diphenylcarbazide solution. The test solution was prepared by combining 200 µl sample under investigation, 400 µl 20 mM MOPS-NaOH buffer, 33 µl 3M H₂SO₄, 40 µl S-diphenylcarbazide solution and 327 µl distilled water. The optical density of the above solution was measured at 540 nm using a spectrophotometer.

A standard curve of Cr⁺⁶ concentration against optical density was prepared in the similar way except that in this case 20 µl of known concentration of K₂Cr₂O₇ was used in place of the supernatant. Using this standard curve the residual Cr⁺⁶ concentration of batch experiments or Cr⁺⁶ concentration of chemostat effluent were determined.

3. THEORETICAL ANALYSIS

3.1 Determination of Kinetic Parameters

Determination of intrinsic growth kinetic parameters, viz., substrate saturation constant, maximum specific growth rate and inhibition constant, if any, can conveniently be done from the analysis of

batch performance data. From the results, reported by earlier investigators, it is understood that the bioconversion reaction of Cr^{+6} to Cr^{+3} is inhibited by the substrate when the former is used at a higher concentration level. Since in the present investigation the range of initial concentration is fairly high (70-90 mg/dm^3) the reaction rate may logically be assumed to be inhibited by the initial Cr^{+6} concentration. Under such situation the most classical semi deterministic model equation proposed by Haldane is still an important tool for prediction of cell growth rate. The classical substrate inhibited Haldane equation for cell growth rate is given by

$$\mu = \frac{\mu_{\max} C_A}{K_S + C_A + \frac{C_A^2}{K_i}} \quad (1)$$

The above equation contains three kinetic parameters, viz., μ_{\max} , K_S , K_i . More over, the concentration of substrate at which the maximum growth of microorganisms occurs ($C_{A_{\max}}$) is also an important parameter for growth kinetic study. Shuler and Kargi (Shuler and Kargi, 1992) have given an expression for obtaining the magnitude of $C_{A_{\max}}$ from the known value of kinetic parameters as follows

$$C_A = \sqrt{K_S K_i} = C_{A_{\max}} \quad (2)$$

Eq. (1) clearly shows that the specific growth rate is strongly dependent upon the initial substrate concentration and a plot of μ vs C_A can be used to identify the substrate uninhibited regime and substrate inhibited regime. The point of inflexion of the curve will indicate the substrate concentration for maximum growth rate ($C_{A_{\max}}$). The data lying in the region of substrate uninhibited section can be used to determine μ_{\max} and K_S from the double reciprocal plots of $1/\mu$ against $1/C_A$. On the other hand experimental data lying in the substrate inhibited region can be used to determine the substrate inhibition constant K_i from the plot $1/\mu$ against C_A . In the present investigation the above exercise has been meticulously done and the magnitude of each kinetic parameter has been determined. These are shown in Table 1.

3.2 Model Equation for Chemostat

The mathematical model equation for predicting the substrate concentration in the reactor effluent has been carried out using following assumptions:

1. Microorganisms follow the same intrinsic growth kinetics.
2. The density of reaction mixture is reasonably constant.
3. Ideal mixing condition exists in the Chemostat.

The schematic of the flow pattern in the chemostat used in the present investigation for the transformation of Cr^{+6} to Cr^{+3} is shown in Fig. 1.

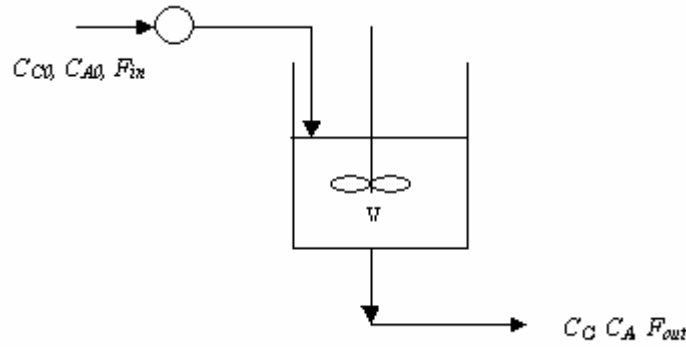


Figure 1. Schematic representation of chemostat

The material balance equation for substrate in chemostat of constant reaction volume under dynamic condition may be written as:

$$V \frac{dC_A}{dt} = F_{in} C_{A_0} - F_{out} C_A - \frac{1}{Y_{C/A}} \mu C_C V \quad (3)$$

Under the steady state condition when the Eq. (3) becomes:

$$D C_{A_0} - D C_A - \frac{1}{Y_{C/A}} \mu C_C = 0 \quad (4)$$

$$\text{where } D = \frac{F}{V} \text{ and } F_{in} = F_{out} = F \quad (5)$$

Similarly the differential mass balance equation for biomass under dynamic state is given by:

$$\frac{dC_C}{dt} = -D C_{C_0} + \mu C_C \quad (6)$$

Under steady state condition $\frac{dC_C}{dt} = 0$, giving an important relation between the specific growth rate and the dilution rate as follows:

$$D = \mu \quad (7)$$

Comparing Eq. (1) and Eq. (7) one gets:

$$D = \frac{\mu_{\max} C_A}{K_S + C_A + \frac{C_A^2}{K_i}} \quad (8)$$

A simple calculation on Eq. (8) gives a simulated equation for C_A as follows:

$$C_A = \frac{-(DK_i - K_i\mu_{\max}) - \sqrt{(DK_i - K_i\mu_{\max})^2 - 4D^2K_iK_S}}{2D} \quad (9)$$

Thus knowing the kinetic parameters and form a preset value of dilution rate one can predict the chromium concentration in the reactor effluent and consequently one can calculate the extent of conversion as well.

4. RESULTS AND DISCUSSION

The cell growth time history curves and Cr^{+6} depletion as a function of initial Cr^{+6} concentration, derived from batch mode reaction, are shown in Fig.2 and Fig.3 for *Pseudomonas* sp. and *Bacillus* sp. respectively. The simulated values of the above variables for both the above mentioned monocultures have been computed using Haldane equation and have been plotted on the same figure. The figures clearly indicate the presence of an insignificant lag period followed by the usual exponential phase. Lag phase is not very clear probably due to prior adaptation of the microbes during purification stage. Analysis of substrate depletion curves reveals that at higher Cr^{+6} concentration, the trend of the exponential decay is not followed. This nature is an indication of substrate inhibition of cell growth at higher initial Cr^{+6} concentration as observed also by other investigators.

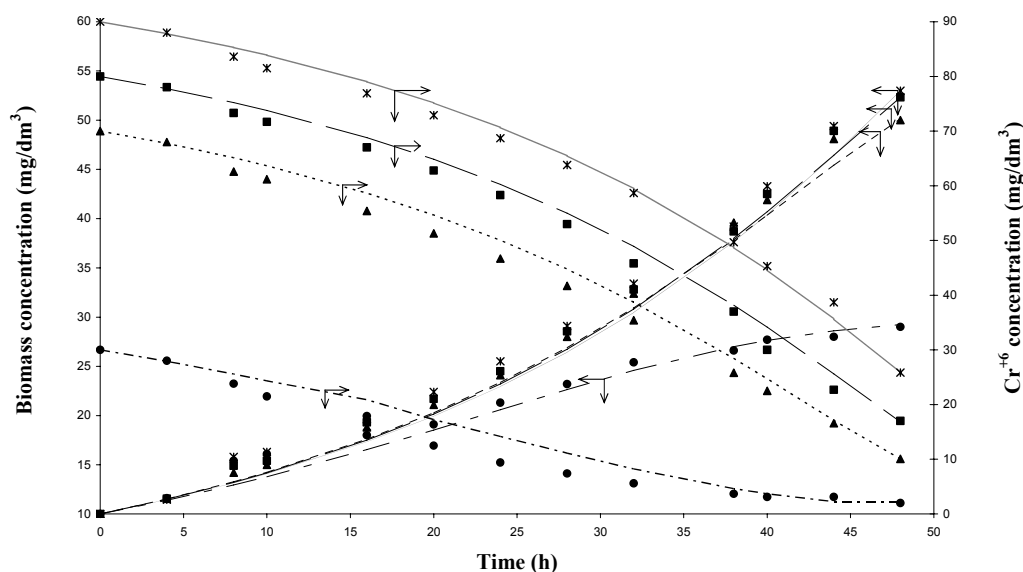


Figure 2. Time history of hexavalent chromium and biomass (*Pseudomonas* sp.) concentration with initial Cr^{+6} concentration as a parameter. (* 90mg/dm³ Experimental; ■ 80mg/dm³ Experimental; ▲ 70mg/dm³ Experimental; ● 30mg/dm³ Experimental; — 90mg/dm³ Simulated; - - - 80mg/dm³ Simulated; 70mg/dm³ Simulated; - 30mg/dm³ Simulated)

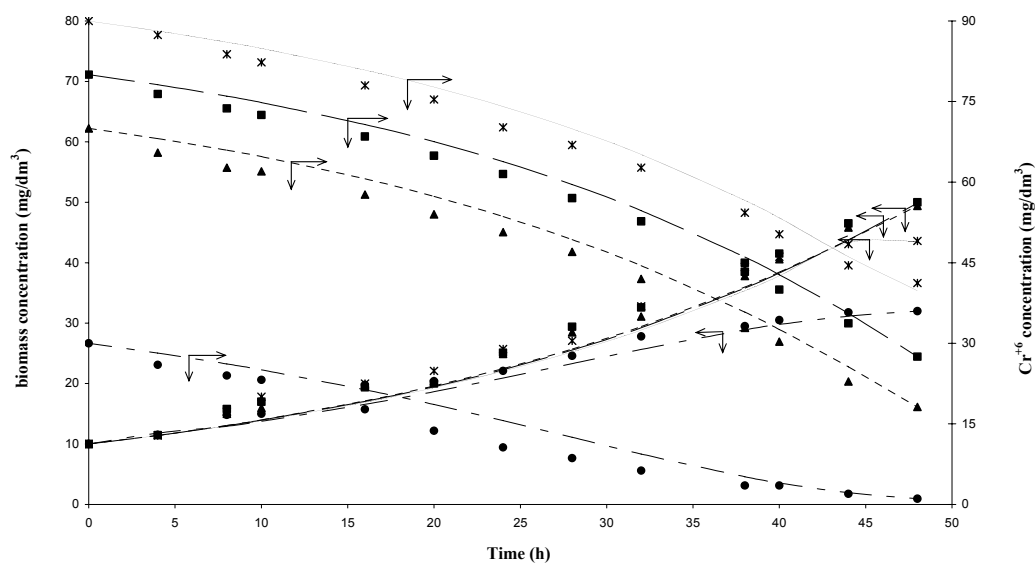


Figure 3. Time history of hexavalent chromium and biomass (*Bacillus* sp.) concentration with initial Cr^{+6} concentration as a parameter. (* 90mg/dm³ Experimental; ■ 80mg/dm³ Experimental; ▲ 70mg/dm³ Experimental; ● 30mg/dm³ Experimental; — 90mg/dm³ Simulated; --- 80mg/dm³ Simulated; -.-.- 70mg/dm³ Simulated; 30mg/dm³ Simulated)

The initial specific cell growth rate (μ) has been computed and plotted against the corresponding initial Cr^{+6} concentration, which is shown in Fig.4. Both the figures shown in Fig.4 show that μ increases up to a particular Cr^{+6} concentration, beyond which the value of μ declines with increasing initial Cr^{+6} concentration. This is the clear indication of the presence of substrate inhibition. In such case, Haldane substrate inhibited model equation has been proposed and different kinetic parameters have been determined following the technique, discussed under theoretical analysis section.

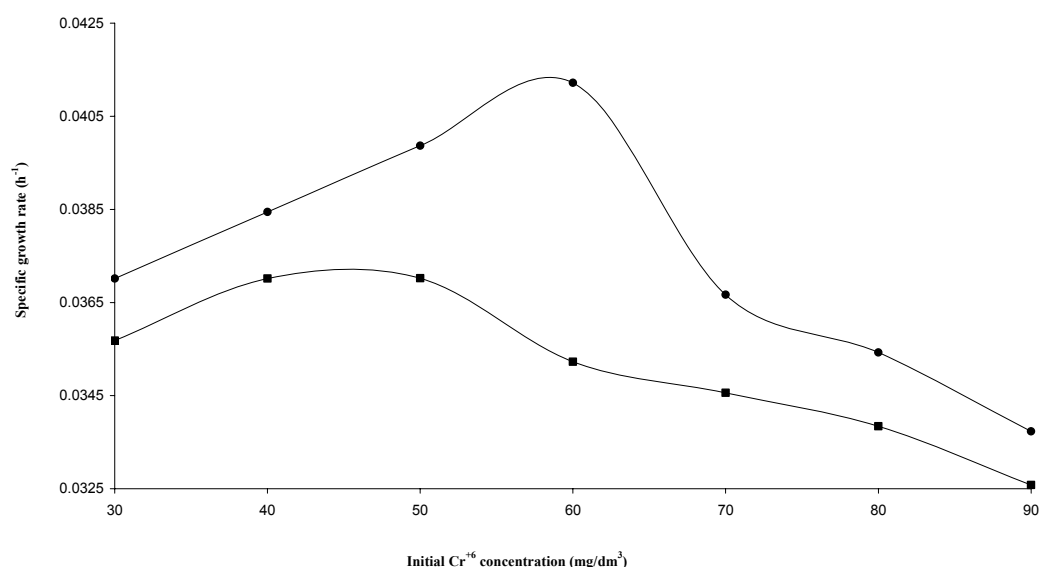


Figure 4. Dependence of specific growth rate of *Pseudomonas* sp and *Bacillus* sp. on hexavalent chromium. (● *Pseudomonas* sp.; ■ *Bacillus* sp)

Experiment on chemostat as the contacting device for the bioconversion of Cr^{+6} to Cr^{+3} has been conducted under steady state condition by varying initial Cr^{+6} concentration from 70 – 90 mg/dm³ and

by setting a fixed dilution rate by adjusting suitable volumetric flow rate. For each fixed dilution rate and for a particular inlet substrate concentration the reactor effluent has been analysed for its Cr^{+6} concentration. The percentage conversions computed from the knowledge of initial substrate concentration have been plotted against the corresponding dilution rate for both the monoculture, *Pseudomonas* sp. and *Bacillus* sp. using Cr^{+6} concentration as parameter. These are shown in Fig.5 and Fig.6.

As expected the conversion of Cr^{+6} increases with the decreasing dilution rate due to longer residence time of the reactant inside the reactor. The simulated value of Cr^{+6} concentration has been calculated using Eq.(9) and the same has been plotted on Fig. 5 and Fig. 6 for comparison. It is observed that the experimental data fit reasonably well with the simulated values. It may therefore be concluded that the proposed model equation from computation of C_A and C_C as given in Eq.(9) and Eq.(4) respectively can conveniently be used for predicting the substrate conversion and the corresponding biomass concentration. From the analysis of Fig.5 and Fig. 6 it is observed that the maximum Cr^{+6} reduction takes place at dilution rate 0.029 h^{-1} for *Pseudomonas* sp. (77.21%) and *Bacillus* sp. (79.22 %).

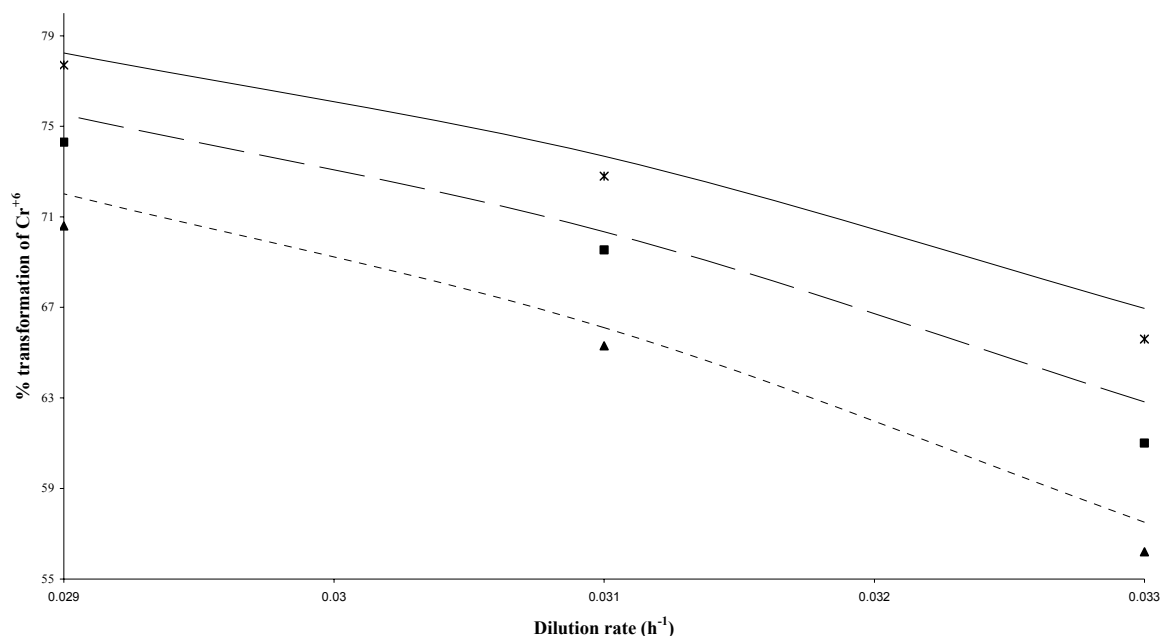


Figure 5. Dependence of percentage transformation of Cr^{+6} on dilution using *Pseudomonas* sp. (* 90mg/dm³ Experimental; ■ 80mg/dm³ Experimental; ▲ 70mg/dm³ Experimental; _____ 90mg/dm³ Simulated; - - - - 80mg/dm³ Simulated; 70mg/dm³ Simulated)

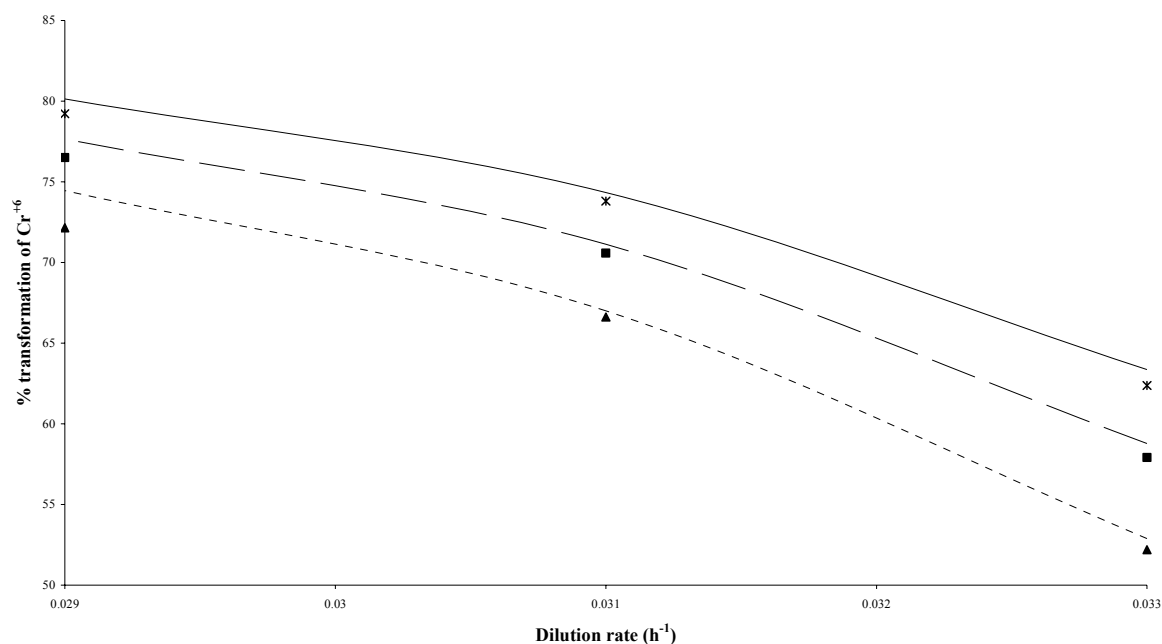


Figure 6. Dependence of percentage transformation of Cr^{+6} on dilution using *Bacillus* sp. (* 90mg/dm³ Experimental; ■ 80mg/dm³ Experimental; ▲ 70mg/dm³ Experimental; _____ 90mg/dm³ Simulated; - - - - 80mg/dm³ Simulated; 70mg/dm³ Simulated)

5. CONCLUSION

Studies on bioconversion of Cr^{+6} to Cr^{+3} have been conducted using *Pseudomonas* sp. and *Bacillus* sp. separately. Initially the microorganisms have been isolated from the local tannery effluent and later have been purified and identified regarding their genera. Growth kinetics of each of the above microorganisms has been studied using a batch fermentor and the information obtained from this experiment has been utilized to determine the kinetic parameters using Haldane type substrate inhibited model equation. The process engineering study has been extended to a chemostat with an intention to predict the reactor performance. In the present investigation, a judicious simulation work has been carried out to develop semi-deterministic model equation for predicting Cr^{+6} concentration in the reactor content as well as the corresponding biomass concentration. Comparison of experimental data with the simulated values indicates the validity of the simulated equation.

Table 1. Growth kinetic parameters of two different bacterial cultures

| Microbial strain | Mmax (hr-1) | KS (mg/dm3) | Ki (mg/dm3) | Camax (mg/dm3) | YC/S |
|------------------------|-------------|-------------|-------------|----------------|-------|
| <i>Pseudomonas</i> sp. | 0.046 | 16.32 | 200.37 | 57.18 | 0.665 |
| <i>Bacillus</i> sp. | 0.056 | 9.43 | 305.29 | 53.66 | 0.776 |

ACKNOWLEDGEMENTS

Authors gratefully acknowledge the financial support received from the Department of Science and Technology, Govt. of India to carry out this research work.

NOMENCLATURE

Cr^{+3} Trivalent chromium

| | |
|------------------|---|
| Cr^{+6} | Hexavalent chromium |
| C_C | Biomass concentration, mg/dm^3 |
| C_A | Substrate concentration, mg/dm^3 |
| $C_{A_{\max}}$ | Substrate concentration at which the maximum growth of microorganisms occurs mg/dm^3 |
| $Y_{C/A}$ | Yield coefficient of cell mass with respect to substrate concentration = Mass of cells produced / mass of substrate consumed |
| K_S | Monod constant, mg/dm^3 |
| K_i | Inhibition constant |
| V | Working volume of Reactor, dm^3 |
| F | Flow rate, ml/h |
| F_{in} | Inlet flow rate, ml/h |
| F_{out} | Outlet flow rate, ml/h |
| D | Dilution rate |
| μ | Specific cell growth rate, h^{-1} |
| μ_{\max} | Maximum specific cell growth rate, h^{-1} |

REFERENCES

- Arellano, H.G., Alcalde, M. and Ballesteros, A. 2004. Use and improvement of microbial redox enzyme for environmental purposes. *Microbial Cell Factories*. 3(10), doi: 10.1186/1475-2859-3-10.
- Bajt, S., Clark, S.B., Sutton, S.R., Rivers, M.L. and Smith, J.V. 1993. Synchrotron x-ray microprobe determination of chromate content using x-ray absorption near-edge structure. *Anal. Chem.* 65, 800-1804.
- EPA (Environmental Protection Agency). 1998. Toxicological Review of Hexavalent Chromium. US Environmental Protection Agency, Washington DC.
- Flores, A., Perez, J.M. 1999. Cytotoxicity, apoptosis, and in vitro DNA damaged induced by potassium chromate. *Toxicol Appl Pharmacol.* 161, 75-81.
- Ganguly, A., Tripathi, A.K. 2002. Bioremediation of toxic chromium from electroplating effluent by chromate-reducing *Pseudomonas aeruginosa* A2Chr in two bioreactors. *Appl Microbiol Biotechnol* 58, 416-420.
- Gu, J.D. and Cheung, K.H. 2001. Phenotypic expression of *Vogesella indigofera* upon exposure to hexavalent chromium, Cr^{6+} . *World Journal of Microbiology and Biotechnology*. 17, 475-480.
- Losi, M.E., Amrhein, C. and Frankenberger, W.T. 1994. Environmental biochemistry of chromium. *Rev Environ Contam Toxicol.* 136, 91-121.
- Lurie, P. and Wolfe, M.S. 2002. Continuing exposure to hexavalent chromium, a known lung carcinogen: An analysis of OSHA compliance inspections. *American journal of Industrial Medicine.* 5, 378-383.
- McLean, J. and Beveridge, T.J. 2000. Chromate reduction by a Pseudomonad isolated from a site contaminated with chromate copper arsenate. *Applied and Environmental Microbiology.* 2, 611-619.
- Middleton, S.S., Latmani, R.B., Mackey, M.R., Ellisman, M.H., Tebo, B.M. and Criddle, C.S. 2003. Cometabolism of Cr(VI) by *Shewanella oneidensis* MR-1 Produces Cell-Associated Reduced Chromium and Inhibits Growth. Wiley InterScience(www.interscience.wiley.com) doi: 10.1002/bit.10725.
- O'Brien, Xu, J. and Patierno, S.R. 2001. Effect of glutathione on chromium-induced DNA crosslinking and DNA polymerase arrest. *Molecular and Cellular Biochemistry.* 222, 173-182.
- Pattanapitpaisal, P., Brown, N.L. and Macaskie, L.E. 2001. Chromate reduction and 16S rRNA identification of bacteria isolated from a Cr(VI)-contaminated site. *Appl Microbiol Biotechnol.* 57, 257-261.
- Pattanapitpaisal, P., Brown, N.L. and Macaskie, L.E. 2001. Chromate reduction by *Microbacterium liquefaciens* immobilized in polyvinyl alcohol. *Biotechnology Letters* 23, 61-65.
- Shuler, M.L. and Kargi, F. 1992. How cells grow. In: *Bioprocess Engineering Basic Concepts*, pp 169-170, 151. (Acrivos, A., Dahler, J., Hanratty, T.J., Prausnitz, J.M. and Scriven, L.E. Eds.). Prentice-Hall, Inc., New Jersey Press.
- Stern, R.M. 1982. Chromium compounds: production and occupational exposure. In: *Biological and Environmental Aspects of Chromium*, pp. 547. (Langard, S., Ed.). Amsterdam: Elsevier Press.
- Stearns, D.M., Kennedy, L.J., Courtney, K.D., Giangrande, P.H., Phieffer, L.S. and Wetterhahn, K.E. 1995. Reduction of chromium (VI) by ascorbate leads to chromium-DNA binding and DNA strand breaks in vitro. *Biochemistry.* 34, 910-919.
- Turick, C. E. and Apel, W.A. 1997. A bioprocess strategy that allows for the selection of Cr (VI)-reducing bacteria from soils. *Journal of industrial Microbiology & Biotechnology.* 18, 247-250.
- Venitt, S. and Levy, L.S. 1974. Mutagenicity of chromates in bacteria and its relevance to chromate carcinogenesis. *Nature.* 250, 493-495.

Zhang, Z., Leonard, S.S., Wang, S., Vallyathan, V., Castranova, V., and Shi, X. 2001. Cr(VI) induces cell growth arrest through hydrogen peroxide. *Molecular and Cellular Biochemistry* 1(2), 77-83.